# Technical Data Sheet

# PE-Cy<sup>™</sup>7 Mouse anti-Human CD279 (PD-1)

# **Product Information**

**Material Number:** 561272

hPD-1; PD1; PDCD1; Programmed cell death 1; SLEB2 Alternate Name:

Size Vol. per Test: 5 μ1 EH12.1 Clone:

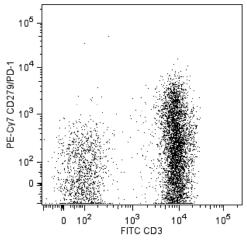
Human PD-1 Recombinant Protein Immunogen:

Isotype: Mouse IgG1, κ Reactivity: QC Testing: Human

Storage Buffer: Aqueous buffered solution containing BSA and ≤0.09% sodium azide.

## Description

The EH12.1 monoclonal antibody specifically binds to CD279. CD279 is an immunoregulatory receptor that is expressed on activated T cells, B cells and myeloid cells and contains an immunoreceptor tyrosine-based inhibitory motif (ITIM) in the cytoplasmic region. Mice deficient in CD279 show a breakdown of peripheral tolerance and manifest multiple autoimmune symptoms. PD-L1 and PD-L2 are ligands of CD279 and are members of the B7 gene family. Interaction of CD279:PD-Ligands results in inhibition of T cell proliferation and cytokine secretion. Reports suggest that the B7/CTLA-4 pathway functions primarily to attenuate, limit, and/or terminate naïve T-cell activation in secondary lymphoid organs. The PD-ligand: CD279 pathway, on the other hand, may primarily attenuate, limit, and/or terminate T-, B-, and myeloid cell activation/effector function at sites of inflammation in the periphery.



Flow cytometric analysis of CD279 (PD-1) expression on human peripheral blood lymphocytes. Whole blood was stained with PE-Cy™7 Mouse anti-Human CD279 (PD-1) (Cat. No. 561272) and FITC Mouse Anti-Human CD3 (Cat. No. 555332) antibodies. The erythrocytes were lysed with BD PharmLyse™ Lysing Buffer (Cat. No. 555899). The two-color flow cytometric dot plot showing the correlated expression patterns of CD3 versus CD279/PD-1 were derived from gated events with the forward and side light-scatter characteristics of viable lymphocytes. Flow cytometry was performed on a BD LSR™ II Flow Cytometry

## **Preparation and Storage**

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.

The antibody was conjugated with PE-Cy7 under optimum conditions, and unconjugated antibody and free PE-Cy7 were removed. Store undiluted at 4°C and protected from prolonged exposure to light. Do not freeze.

### **Application Notes**

# Application

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Flow cytometry	Routinely Tested	

# **Suggested Companion Products**

Catalog Number	Name	Size	Clone	
555899	Lysing Buffer	100 ml	(none)	
555332	FITC Mouse Anti-Human CD3	100 tests	UCHT1	
554656	Stain Buffer (FBS)	500 ml	(none)	

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#### **Product Notices**

- 1. This reagent has been pre-diluted for use at the recommended Volume per Test. We typically use 1 × 10<sup>6</sup> cells in a 100-μl experimental sample (a test).
- 2. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
- 3. PE-Cy7 is a tandem fluorochrome composed of R-phycoerythrin (PE), which is excited by 488-nm light and serves as an energy donor, coupled to the cyanine dye Cy7, which acts as an energy acceptor and fluoresces maximally at 780 nm. PE-Cy7 tandem fluorochrome emission is collected in a detector for fluorescence wavelengths of 750 nm and higher. Although every effort is made to minimize the lot-to-lot variation in the efficiency of the fluorochrome energy transfer, differences in the residual emission from PE may be observed. Therefore, we recommend that individual compensation controls be performed for every PE-Cy7 conjugate. PE-Cy7 is optimized for use with a single argon ion laser emitting 488-nm light, and there is no significant overlap between PE-Cy7 and FITC emission spectra. When using dual-laser cytometers, which may directly excite both PE and Cy7, we recommend the use of cross-beam compensation during data acquisition or software compensation during data analysis.
- 4. Please observe the following precautions: Absorption of visible light can significantly alter the energy transfer occurring in any tandem fluorochrome conjugate; therefore, we recommend that special precautions be taken (such as wrapping vials, tubes, or racks in aluminum foil) to prevent exposure of conjugated reagents, including cells stained with those reagents, to room illumination.
- 5. Warning: Some APC-Cy7 and PE-Cy7 conjugates show changes in their emission spectrum with prolonged exposure to formaldehyde. If you are unable to analyze fixed samples within four hours, we recommend that you use BD<sup>TM</sup> Stabilizing Fixative (Cat. No. 338036).
- 6. For fluorochrome spectra and suitable instrument settings, please refer to our Fluorochrome Web Page at www.bdbiosciences.com/colors.
- 7. Cy is a trademark of Amersham Biosciences Limited. This conjugated product is sold under license to the following patents: US Patent Nos. 5,486,616; 5,569,587; 5,569,766; 5,627,027.
- 8. Caution: Sodium azide yields highly toxic hydrazoic acid under acidic conditions. Dilute azide compounds in running water before discarding to avoid accumulation of potentially explosive deposits in plumbing.
- 9. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
- 10. This product is subject to proprietary rights of Amersham Biosciences Corp. and Carnegie Mellon University and made and sold under license from Amersham Biosciences Corp. This product is licensed for sale only for research. It is not licensed for any other use. If you require a commercial license to use this product and do not have one return this material, unopened to BD Biosciences, 10975 Torreyana Rd, San Diego, CA 92121 and any money paid for the material will be refunded.

#### References

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Latchman Y, Wood CR, Chernova T, et al. PD-L2 is a second ligand for PD-1 and inhibits T cell activation. *Nat Immunol.* 2001; 2(3):261-268. (Biology) Nishimura H, Minato N, Nakano T, Honjo T. Immunological studies on PD-1 deficient mice: implication of PD-1 as a negative regulator for B cell responses. *Int Immunol.* 1998; 10(10):1563-1572. (Biology)

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